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Evidence of Osteolathyrism Among Patients Suffering From Neurolathyrism in Bangladesh

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ABSTRACT In a study of 500 patients suffering from neurolathyrism in Bangladesh it was found that 60 (all male) complained of bone pain and showed skeletal deformities suggestive of osteolathyrism. On X-ray examination a failure of fusion in both vertebral and iliac epiphyses was found in two patients. At the age of these patients (30 and 37 years) such failure was considered a clear evidence of osteolathyrism. All 60 patients were accustomed to eating the green parts of *Lathyrus sativus,* which contain 2-cyanoethyl-isoxazolin-5-one, a compound that chemically and metabolically can produce the osteolathyrogen β-aminopropionitrile (BAPN), as well as foods made from the seeds of the same plant which contain the neurotoxin 3-N-oxalyl-2,3-diaminopropanoic acid (β-ODAP). *Nat. Toxins 5:43–46, 1997.* © 1997 Wiley-Liss, Inc.

Key Words: *Lathyrus sativus;* lathyrism; osteolathyrism; neurolathyrism; 2-cyanoethyl-isoxazolin-5-one; β-ODAP; BAPN

INTRODUCTION

Various species of *Lathyrus* are cultivated throughout the world, and it has been known from earliest times that some of these are toxic when eaten by man and domestic animals. Selye [1957] recognised that there were at least two toxic syndromes associated with the ingestion of *Lathyrus* seeds. The first of these, termed neurolathyrism, is the classical disease seen in man which involves impairment of leg muscles in different degrees up to paralysis of the legs. The disease is associated with the ingestion of food made from the seeds of *Lathyrus sativus* (grass pea, chickling pea, khesari, guaya, san lee dow, pois carré) or less frequently from those of *L. cicera* and *L. clymenum.* The second, termed osteolathyrism, is a condition involving skeletal changes and the loss of elasticity of blood vessel walls that occurs in laboratory animals fed on *L. odoratus* (sweet pea).

The first toxin to be identified in a species of the genus was γ-glutamyl-β-aminopropionitrile which was isolated from seeds of *L. pusillus* [Dupuy and Lee, 1954] and *L. odoratus* [McKay et al., 1954] and characterised by Schilling and Strong [1954]. The compound proved to be an osteolathyrogen, the active portion of the molecule being β-aminopropionitrile (BAPN) [Dasler, 1954]. A second compound which was acutely toxic to rats was subsequently isolated from seeds of *L. latifolius* and identified by Ressler et al. [1964] as L-2,4-diaminobutanoic acid (DABA). A survey of compounds present in the seeds of 52 species of *Lathyrus* [Bell, 1962, 1964] showed however that the three species implicated as causes of human lathyrism did not contain BAPN, its γ-glutamyl derivative or DABA. All three species did however contain a strongly acidic amino acid which was isolated from *L. sativus* by two independent groups [Adiga et al., 1963; Murti et al., 1964] and characterised by Rao et al. [1964] as 3-N-oxalyl-L-2,3-diaminopropanoic acid (β-ODAP) which is also sometimes referred to as β-oxalylaminoalanine (BOAA). β-ODAP is a neurotoxin and without doubt it is the prime cause of neurolathyrism in man.

Despite the absence of BAPN and its γ-glutamyl derivative from the seeds of *L. sativus* it has been reported that minor skeletal lesions suggestive of osteolathyrism occur in some older patients [Weintroub et al., 1980; Cohn and Streifler, 1981, Cohn, 1986]. It was suggested [Lambein et al., 1993] that the presence of low concentrations of 2-cyanoethyl-isoxazolin-5-one (compound VIII) in the seeds might be responsible. This heterocyclic compound, which can be photolysed [De Bruyn et al., 1992] or metabolised to BAPN, produces the symptoms of osteolathyrism in rats [Van Rompuy et al., 1974] and chicks [Lambein and De Vos, 1981].

In this paper we present further evidence that osteolathyrism is not merely a syndrome that can be induced experimentally in laboratory animals but a condition that affects human populations.

MATERIAL AND METHODS

During 1991 and 1992 a survey was made of 500 patients suffering from neurolathyrism in three districts of Northwest

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Bangladesh. Identification of neurolathyrism was done by a method of exclusion from some 2000 neurological patients as described elsewhere [Haque et al., 1991; Haque, 1992]. Of these, 60 (all male) were selected for radiological examination on the basis of the following criteria: bowing of the legs at the knee joints, kyphoscoliosis, backache and the onset of neurolathyrism before the age of 17 (i.e., *L. sativus* had been part of their diet before the age at which the ossification centres would normally be expected to unite).

At the time of examination all 60 patients were above the age of 30 (mean age 37 $\pm$ 2.4 years). The mean age at onset of neurolathyrism for the group was 13 $\pm$ 2 years. All had consumed green leaves and/or seedlings as well as food made from the seeds of *L. sativus* prior to the onset of the disease.

The long bones, vertebral bodies and pelvic bones of each patient were X-rayed. In two patients there was a clear evidence of a failure of fusion of the vertebral and iliac epiphyses (Figs. 1 and 2).

Case Reports

Case 1 (examined 11th May, 1992)

A right-handed male labourer aged 37 who was healthy until the age of 17. Presently engaged in house work, previously on the land. Onset of the disease was sudden, after working in the field for about two hours in a sitting position he suffered spasms in the muscles of both legs when he stood up. A gust of wind caused him to fall. When he rose again he had difficulty in walking and at night he felt mild pain with spasms in the muscles of both legs. The following morning he could not leave his bed nor stand unaided. Ever since, he has suffered weakness and heaviness in both legs. Muscle spasms have also continued, mainly at night. He has experienced an urgency of micturition for 18 years. His knee joints, ankles, and toes became deformed over a period of 3 years following the onset of neurolathyrism. Those deformities are still present. He suffers low back pain over lumbosacral vertebrae and pain in the muscles of both legs after minimal activity. No fever, trauma, or other symptoms were reported. His pulse was 80/min and his blood pressure 100/70 mm Hg.

The patient had normal higher cerebral functions, cranial nerves, and sensory system. The motor system showed disuse atrophy of the small muscles of the legs without fasciculation. The power of both upper limbs was normal while the lower limbs had grade 4 weakness, also the tone was increased in both lower limbs. Reflexes were increased in the lower limbs, showing extensor plantar reflexes with ankle clonus. Coordination of movements was normal except for the typical spastic scissor gait.

The patient had no past illnesses that might have been contributory to the symptoms, and he had three brothers and two sisters who were unaffected. The patient was a low income day labourer who smoked 15–20 cigarettes per day.

Fig. 1. X-ray of the vertebrae of a 37-year-old neurolathyrism patient with evidence of osteolathyrism. The arrow shows lack of union of ossification centre of vertebrae.

As medication he was given vitamin B complex during the early stages of the disease, with no significant effect. He took about 500 g of *L. sativus* seeds per day regularly in the form of chapatis or soup continously for one year prior to the onset of the disease. He also took the green leaves of *L. sativus* regularly when in season. After the onset of the disease he took food made from the seeds occasionally and green leaves when in season. Otherwise his food consisted of three meals (high carbohydrate) each day and animal protein only once or twice a month.

A clinical diagnosis of neurolathyrism was made with radiological evidence of osteolathyrism.

Case 2 (examined 26th February, 1992)

A right-handed male farmer aged 30 years. Healthy until the age of 10 years. Gradual weakness and heaviness of lower limbs at onset increasing over 2–3 months by which time his toes were scraping the ground during walking. The spasms of the leg muscles also appeared during this period. Deformity of both his lower limbs (knees, ankles, and toes)

Fig. 2. X-ray of the hip of a 30-year-old neurolathyrism patient with evidence of osteolathyrism. Arrow shows lack of union of ossification centre.

became apparent 4 years after the onset of the neurological symptoms and maximum deformity was reached by the age of 20 years. Urgency of micturition during the last 12 years, now unable to hold urine for more than 5 min after first signal of micturition. No fever, trauma, or other symptoms reported.

The patient had normal pulse and blood pressure. The patient had normal higher cerebral functions and sensory system, the cranial nerves were normal except for congenital drooping of the upper left eyelid. His motor system was normal generally, with normal power in the upper limbs, but grade 4 weakness in both lower limbs. Also the tone was increased only in the lower limbs. All deep tendon reflexes were increased in both lower limbs with ankle clonus and extensor plantar reflexes, but the overall coordination of movements was normal.

The patient had no past illnesses that might have been contributory to the symptoms. His father suffered from neurolathyrism but one brother and one sister were unaffected. The patient was a farmer cultivating his own land, having an income five times greater than of case 1. He took vitamin B complex during the early stages of the disease and still occasionally takes Berin (B1) and Cytamine (B12) injections. After taking B1 he feels better.

The patient had eaten the seeds and leaves (when in season) of *L. sativus* for a prolonged period before the onset of the disease. He still occasionally takes the seeds as soup and eats the green leaves when available.

A clinical diagnosis of neurolathyrism was made with radiological evidence of osteolathyrism.

DISCUSSION

In the past osteolathyrism has been regarded exclusively as an experimentally induced condition found in laboratory animals that have been fed on the seeds of *L. odoratus* (sweet pea). These seeds accumulate β-aminopropionitrile (BAPN) in the form of its γ-glutamyl derivative and BAPN inhibits cross linking between polypeptide chains during the synthesis of both collagen and elastin. The failure to form such links causes skeletal deformation and blood vessel fragility in experimental animals [Levene, 1962; O'Dell et al., 1966]. The inhibition of cross linking is dose dependent [Lee et al., 1990] and also results in a decrease in the elastic modulus of the bone [Lees et al., 1987].

Despite the absence of BAPN and its γ-glutamyl derivative from seeds of *L. sativus*, bone deformities were found on X-ray films of older patients suffering from chronic neurolathyrism since 30 years. Weintroub and coworkers [1980] reported abnormal skeletal findings in the roentgenological files from 3 of 23 neurolathyrism patients. Cohn [1986] reported skeletal lesions indicative of osteolathyrism in 6 of 55 patients suffering from neurolathyrism. In both cases the patients had been prisoners in a forced labour camp during the second world war when their daily rations were 400 g of *L. sativus* seed cooked in water and 200 g of bread made from barley and chopped straw (20%).

In comparison to those patients discribed by Weintroub et al. [1980] and by Cohn [1986], the Bangladeshi patients described in this paper contracted neurological symptoms at an earlier age and also consumed the green parts of the plant

besides the seeds. It has been suggested [Lambein et al., 1993] that low concentrations of 2-cyanoethyl-isoxazolin-5-one (compound VIII) in the seeds could have caused the osteolathyrism. Our present findings show that osteolathyrism also occurs among neurolathyritic patients in Bangladesh. A significant difference between the two groups of patients is the practice of those in Bangladesh of eating not only the seeds, but also the young seedlings and the fresh green shoots of the plant as a vegetable. Especially the seedlings contain significant concentrations of compound VIII as well as β-ODAP and a second neurotoxin α-amino-γ-(isoxazolin-5-on-2-yl)-butanoic acid (compound VI) [Lambein et al., 1992a, 1993]. In animals compounds VI and VIII can be metabolized with formation of the toxins DABA and BAPN, respectively [Lambein and De Vos, 1981; Van Rompuy et al., 1974], and these heterocyclic compounds were suggested to be the precursors for those toxins in *Lathyrus* species [Lambein et al., 1992b]. We suggest that those who eat the green parts of the plant and especially the young seedlings could well be at greater risk of developing osteolathyrism than those who only eat foods made from the seeds.

While the number of patients showing skeletal changes suggestive of osteolathyrism was only 12% of the total number examined, the significance of these findings cannot be ignored nor can the possibility that some individuals at risk may develop osteolathyrism without showing symptoms of neurolathyrism. This possibility has not yet been investigated.

The present findings emphasise the need to determine the concentrations of osteolathyrogens as well as neurolathyrogens in the seeds and green parts of established and newly developed varieties of *Lathyrus sativus* and the foods prepared from them. The intake of the various compounds needed to produce toxic effects in man must also be established if the potential risks associated with the use of this valuable plant are to be avoided.

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